B.7.7.3 Magnitude of Residues on Set of Representative Processes (Annex IIA 6.5.3; Annex IIIA 8.5.3)

B.7.7.3.1 Sunflower

Document ID:	MRID No. 50489805
Report:	Dorshner, K. (2017) "Sulfoxaflor: Magnitude of the Residue on Sunflower." IR-4 PR No. 11095. Laboratory Number 11095.13-FLR 11. Unpublished study prepared by IR-4 project. Rutgers, The State University of NJ. North Princeton, NJ, USA. 302 pages.
Guidelines:	EPA OCSPP Harmonized Test Guideline 860.1520 Processed Food/Feed (August 1996) PMRA Regulatory Directive DIR98-02 – Residue Chemistry Guidelines, Section 10 – Processed Food/Feed OECD Guideline 508 Magnitude of the Pesticide Residues in Processed Commodities (October 2008)
GLP Compliance:	No deviations from regulatory requirements were reported which would have an impact on the validity of the study.
Acceptability:	The study is considered scientifically acceptable.
Evaluator:	Jack Giordano, Chemist, RAB2/HED (7509P)

EXECUTIVE SUMMARY

A processing study with sulfoxaflor on sunflower field trials was conducted in the United States during the 2013 growing season. Sulfoxaflor, formulated as a water dispersible granule (WG), with produce name Transform WG; containing 50% active ingredient (ai), was applied to sunflowers as two foliar broadcast applications at rates of 0.088-0.090 pounds of ai per acre (lbs ai/A), equal to 98.6-100.9 grams of ai per acre (g ai/ha). The re-treatment interval (RTI) was seven days, and the total application rate was 0.178 lbs ai/A (199.5 g ai/ha). Samples of sunflower whole seeds (kernel plus hull) were harvested 14 days after final treatment. The treated and untreated raw agricultural commodities (RACs) used in the processing study were collected from the same plot as was used in magnitude of the residue study 11095.13-ND13. Therefore, the application rate of sulfoxaflor for the processing study was approximately 1X the maximum label rate. The sunflower whole seeds were processed into meal and refined oil using simulated commercial practices.

All samples were frozen at the testing facility and remained frozen during shipping and storage prior to processing and analysis. The maximum storage duration for samples was 115 days (~4 months) from harvest to processing and 710 days (~23.5 months) from processing to analysis. Storage conditions and durations are supported by studies showing that residues of sulfoxaflor, X11719474, and X11721061 are stable in sunflower seed meal for intervals of up to 685 days (~23 months) and refined oil for up to 696 days (~23 months) under frozen conditions.

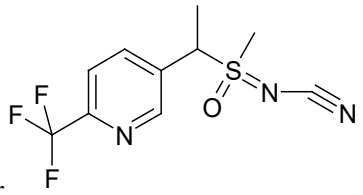
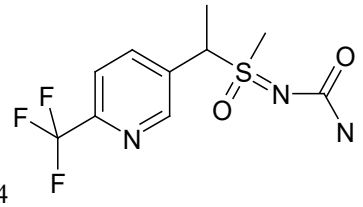
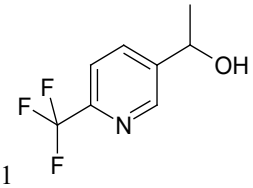
Samples in the current study were analyzed using Method 091116, an LC/MS/MS method to determine residues of sulfoxaflor, X11719474, and X11721601. Acceptable method validation and concurrent recoveries were reported for meal and refined oil samples at fortification levels of

0.01-5.0 mg/kg (ppm), thus validating the method. The limit of quantitation (LOQ), based on the lowest level of method validation (LLMV), was 0.01 ppm per analyte for sulfoxaflor, X11719474, and X11721061.

Only parent sulfoxaflor is considered for both tolerance enforcement and risk assessment, expect in drinking water, where both major metabolites are also considered. A comparison of parent sulfoxaflor residues in the RAC with those in each processed fraction resulted in a processing factor of <0.71 for both meal and refined oil. These processing factors are below the theoretical concentration factors.

I. MATERIALS AND METHODS

A. MATERIALS

Table 7.7.3.1-1. Nomenclature for Sulfoxaflor and Metabolites of Interest.	
Common name	 Sulfoxaflor
Identity	*N*-[methyloxido[1-[6-(trifluoromethyl)-3-pyridinyl]ethyl]-λ^4-sulfanylidenelcyanamide
CAS no.	946578-00-3
Company experimental name	XDE-208 (Dow Agro) ASF 1069 (Syngenta)
Metabolite	 X11719474
Identity	N-((methyl)oxido)(1-[6-(trifluoromethyl)pyridine-3-yl]ethyl)-λ^6-sulfanylidene)urea
Metabolite	 X11721061
Identity	1-[6-trifluoromethyl)pyridine-3-yl]ethanol

B. Study Design

1. Test Procedure

Location and detailed use pattern for the trial is provided in Table B.7.7.3.1-2.

Table B.7.7.3.1-2. Study Use Pattern.							
Location: City, State/Province; Year (Trial ID)[1]	End-use Product/ Formulation (% ai)	Method of Application/ Timing of Application	Volume (gal/A) [L/ha]	Rate per Application (lbs ai/A) [g ai/ha]	Retreatment Interval (days)	Total Rate (lbs ai/A) [g ai/ha]	Surfactant/ Adjuvant
Fargo, ND; 2013 (IDP05)	Transform WG (50%)	1. Foliar broadcast/10% green bracts left	21 [196]	0.090 [100.9]	--	0.178 [199.5]	Prefer 90 NIS
		2. Foliar broadcast/30% brown bracts	21 [196]	0.088 [98.6]	7		

[1] The trial ID number has the prefix 11095.13-

Bulk samples (~51.8 lbs (23.5 kg) each) of untreated and treated sunflower whole seeds were harvested at maturity and transferred to the processing facility for preparation of meal and refined oil. Sunflower whole seeds were processed using simulated industrial practices.

Sample Handling and Preparation

Sunflower heads were cut from throughout the plot, avoiding row ends, and put in seven tote boxes. Samples were fed through a thresher and the seed was collected in dish pans. Samples were wind winnowed by slowly being poured into an empty box. The samples were frozen within 27 minutes of harvest and shipped by ACDS freezer truck to the University of Idaho Food Technology Center. Samples remained frozen and were processed within 105 days (~4 months) of arrival. The samples were shipped to the analytical laboratory (IR-4 Southern Region Laboratory in Gainesville, Florida) by ACDS freezer truck. All samples arrived frozen and intact at the analytical laboratory. The samples were checked in and then stored frozen until extraction and analysis.

Sample Processing

The processing whole seed residue samples were shipped frozen to the processing laboratory by ACDS freezer truck. A representative sample was removed, packaged, labeled and placed in frozen storage. The remaining seed was dried at ~60°C for approximately 30 minutes, followed by screening to remove debris. The seed was dehulled and flaked before heating and expelling to recover crude oil and presscake. The crude oil was degummed and alkali refined to obtain refined oil. Remaining oil in the presscake was removed with hexane to produce meal. The refined oil and meal crop fractions for each sample were then stored frozen.

2. Description of Analytical Procedures

Samples were analyzed for residues of sulfoxaflor, X11719474, and X11721061 using the Analytical Method 091116 titled "Enforcement Method for the Determination of Sulfoxaflor (XDE-208) and its Main Metabolites in Agricultural Commodities using Offline Solid-Phase Extraction and Liquid Chromatography with Tandem Mass Spectrometry Detection." Successful method validation was reported by the registrant and by an independent laboratory validation (ILV).

Minor modifications were made to the reference method to improve performance. Samples were centrifuged for 5 minutes instead of 3 minutes and a 15-mL centrifuge tube was used instead of a

96-well plate. Acetonitrile (ACN) was used as the mixed internal standard solution instead of 90/10 (v/v) methanol/glycerin, and 2.5 mL of sample was added to the centrifuge tube instead of 500 µL. The TurboVap LV evaporator was maintained at 60°C instead of 40°C. The samples were concentrated to about 0.5 mL instead of to near dryness. A 2.5 mL sample of 0.01 N NaOH was added to the centrifuge tube instead of a 500 µL sample, 0.625 mL of 0.25% formic acid was added to the centrifuge tube instead of 0.125 mL, and 1.25 mL of 10 mg/mL glucosidase solution was added to the centrifuge tube instead of 0.250 mL. A Supelclean ENVI-Carb solid-phase extraction (SPE) tube was used instead of an Oasis HLB 96 well-plate. The sample was concentrated to reduce the amount of ACN and brought to the 5 mL mark with 5/95 (v/v) ACN/water and 0.1% formic acid. A different LC/MS/MS model, HPLC column, mobile phase, gradient and injection volume were used. Negative ionization was used for XDE-208 (sulfoxaflor) and its internal standard X11843864 instead of positive ionization. Negative ionization gave increased sensitivity and lower background.

Briefly, samples were extracted with 80/20 (v/v) ACN/water. Meal and refined oil samples were centrifuged. An aliquot of the extract was combined with internal standard solution and evaporated. The extracts requiring dilution were diluted before the addition of internal standard. Aqueous NaOH was added and hydrolyzed at 50°C with glucosidase from Aspergillus Niger solution. The solution was purified with a Supelclean ENVI-Carb SPE cartridge. Extracts were filtered before analysis using LC/MS/MS with heated electro-spray ionization. The LOQ for sunflower meal and refined oil was 0.01 ppm for sulfoxaflor, X11719474, and X11721061, based on the LLMV.

II. RESULTS AND DISCUSSION

Method performance was evaluated during method validation and by use of concurrent recovery samples by fortifying meal and refined oil samples at 0.01 ppm (n=6), 0.50 ppm (n=3) and 5.0 ppm (n=3) for each analyte. All recoveries were within the generally recognized acceptable range of 70% to 120%; therefore, the method was considered valid for the analysis of sulfoxaflor, X11719474, and X11721061 residues in sunflower meal and refined oil matrices (Table B.7.7.3.1-3). The fortification levels did bracket the measured residues.

The detector response was linear (coefficient of determination, r^2 >0.99) within the range of 0.0500 ng/mL to 5.00 ng/mL. Representative chromatograms of control samples, fortified samples, and treated samples were provided. The control chromatograms generally had no peaks of interest above the chromatographic background. The fortified sample chromatograms contained only the analyte of interest, and peaks were symmetrical and well defined.

Table B.7.7.3.1-3. Summary of Method Validation and Concurrent Recoveries of Sulfoxaflor and Metabolites from Sunflower Meal and Refined Oil

Matrix	Fortification Level (ppm)	Recoveries (%)	Mean ± Std. Dev. (%)
Sulfoxaflor			
Meal (Method Validation)	0.01	99, 102, 104	102 ± 2.5
	0.50	86, 89, 87	87 ± 1.5
	5.0	94, 97, 94	95 ± 1.7
Meal (Concurrent Recovery)	0.01	90, 91, 91	91 ± 0.6

Table B.7.7.3.1-3. Summary of Method Validation and Concurrent Recoveries of Sulfoxaflor and Metabolites from Sunflower Meal and Refined Oil			
Matrix	Fortification Level (ppm)	Recoveries (%)	Mean ± Std. Dev. (%)
Refined Oil (Method Validation)	0.01	99, 99, 95	98 ± 2.3
	0.50	85, 85, 85	85 ± 0
	5.0	95, 94, 98	96 ± 2.1
Refined Oil (Concurrent Recovery)	0.01	93, 91, 95	93 ± 2
X11719474			
Meal (Method Validation)	0.01	87, 87, 87	87 ± 0
	0.50	87, 89, 86	87 ± 1.5
	5.0	93, 96, 95	95 ± 1.5
Meal (Concurrent Recovery)	0.01	86, 86, 87	86 ± 0.6
Refined Oil (Method Validation)	0.01	80, 79, 80	80 ± 0.6
	0.50	83, 83, 83	83 ± 0
	5.0	92, 91, 96	93 ± 2.6
Refined Oil (Concurrent Recovery)	0.01	86, 88, 85	86 ± 1.5
X11721061			
Meal (Method Validation)	0.01	88, 91, 89	89 ± 1.5
	0.50	87, 88, 87	87 ± 0.6
	5.0	96, 98, 97	97 ± 1.0
Meal (Concurrent Recovery)	0.01	86, 85, 85	85 ± 0.6
Refined Oil (Method Validation)	0.01	84, 84, 83	84 ± 0.6
	0.50	86, 87, 86	86 ± 0.6
	5.0	95, 94, 98	96 ± 2.1
Refined Oil (Concurrent Recovery)	0.01	84, 85, 89	86 ± 2.6

The maximum storage duration for sunflower meal and refined oil between harvest and extraction for analysis was 705 and 710 days (~23.5 months), respectively (Table B.7.7.3.1-4A). Residues were determined within seven days of extraction.

Freezer storage stability data were generated concurrently with the sunflower meal and refined oil processing study (Table B.7.7.3.1-4B). Sunflower meal and refined oil samples were fortified with 0.1 ppm of sulfoxaflor, X11719474, and X11721061 and stored frozen for 685 and 696 days, respectively. Freezer storage stability recoveries for sunflower meal were 120%, 114%, and 135% for sulfoxaflor, X11719474, and X11721061, respectively. Freezer storage stability recoveries for refined oil were 111%, 100%, and 113% for sulfoxaflor, X11719474, and X11721061, respectively. Although samples were stored for up to 20 days longer than the concurrent storage stability study, given that good recoveries were observed, it is not anticipated that residues would have degraded below acceptable levels during the additional sample storage time. Therefore, it is expected that sulfoxaflor, X11719474, and X11721061 residues were stable in sunflower meal and refined oil under frozen storage for the duration of the storage period.

Table B.7.7.3.1-4A. Summary of Storage Conditions.			
Matrix (RAC or Extract)	Storage Temperature (°C)	Actual Storage Duration (days/months)[1]	Interval of Demonstrated Storage Stability (days/months)
Meal	Generally < -20	705 (~23.5)	A concurrent freezer storage stability study was conducted. The data showed that sulfoxaflor, X11719474, and X11721061 residues are stable when stored frozen in sunflower meal for 685 days (~23 months)
Refined Oil	Generally < -20	710 (~23.5)	A concurrent freezer storage stability study was conducted. The data showed that sulfoxaflor, X11719474, and X11721061 residues are stable when stored frozen in sunflower refined oil for 696 days (~23 months)

[1] From harvest to residue extraction. Residues were determined within seven days of extraction.

Table B.7.6.1.1-4B. Concurrent Freezer Storage Stability Study.						
Matrix	Analyte	Storage Period (days/months)	Fortification Level (ppm)	Freezer Storage Recovery (%) [Average Recovery]	Concurrent Recovery (%)	Corrected Freezer Storage Recovery[1] (%)
Meal	Sulfoxaflor	685 (~23 months)	0.10	136, 113, 110 [120]	89	135
	X11719474	685 (~23 months)	0.10	131, 107, 104 [114]	83	137
	X11721061	685 (~23 months)	0.10	156, 125, 124 [135]	87	155
Refined Oil	Sulfoxaflor	696 (~23 months)	0.10	110, 116, 107 [111]	91	122
	X11719474	696 (~23 months)	0.10	99, 103, 97 [100]	82	122
	X11721061	696 (~23 months)	0.10	116, 116, 108 [113]	89	127

[1] Corrected freezer storage recovery <100% using the following: (Average Freezer Storage Recovery/Concurrent Recovery)*100

Residues found in samples and processing factors are given in Table B.7.7.3.1-5.

Table B.7.7.3.1-5. Residue Data from Sunflower Whole Seed Processing Study with Sulfoxaflor, X11719474, and X11721061.

Commodity	Analyte	Residues (ppm) [Average Residue]	Processing Factor[1]
Whole Seed	Sulfoxaflor	0.015 0.013 [0.014]	--
	X11719474	<0.01	--
	X11721061	<0.01	--
	Total	<0.035 <0.033 [<0.034]	--
Meal	Sulfoxaflor	<0.01	<0.71
	X11719474	<0.01	1
	X11721061	<0.01	1
	Total	<0.03	<0.88
Refined Oil	Sulfoxaflor	<0.01	<0.71
	X11719474	<0.01	1
	X11721061	<0.01	1
	Total	<0.03	<0.88

[1] Factor determined by dividing the residue in processed fraction by residue in RAC in the same trial

III. CONCLUSIONS

The sunflower processing study is considered scientifically acceptable. A comparison of the residues in sunflower whole seeds with those in meal and refined oil fractions indicated that residues of sulfoxaflor, X11719474, and X11721061 do not concentrate in any of the processed commodities. The empirical processing factors are below the theoretical factors. Adequate storage stability data are available to support sample storage durations and conditions.

REFERENCES

PMRA # 1941241. MRID # 47832031. Rodrigues Junior, A. (2010) “Enforcement Method for the Determination of Sulfoxaflor (XDE-208) and its Main Metabolites in Agricultural Commodities using Offline Solid-Phase Extraction and Liquid Chromatography with Tandem Mass Spectrometry Detection.” Laboratory Study ID: 091116. Unpublished study prepared by Dow AgroSciences Ind. Ltda, Mogi Mirim, SP. 93 pages.